NTP Research Concept: Hydroquinone

Project Leader:
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Nomination Background and Rationale:
Hydroquinone (HQ) was nominated by the United States Food and Drug Administration (FDA) for reproductive toxicity and dermal toxicity and carcinogenicity studies to provide data for assessing safety and determining regulatory policy for the chemical in consumer products.

HQ occurs naturally and is produced synthetically for a variety of uses. HQ is present in some foods and beverages and is used as a reducing agent in photographic developing solutions, an antioxidant in rubber and dyes, a polymerization inhibitor in chemicals such as acrylates, a stabilizer in paints, fuels, and oils, a medicine to treat dyschromias (abnormal skin pigmentation), and in cosmetics, including topical preparations used for skin-lightening. HQ inhibits tyrosinase-catalyzed melanin production and selectively damages melanocytes. In rodents and humans, HQ is absorbed following oral or dermal administration and is primarily excreted in urine as glucuronide or sulfate conjugates. A bioactivation pathway for HQ involves formation of benzoquinone and subsequent reaction with sulfhydryl groups. Neurotoxicity has been observed in rodents receiving high oral doses and in humans accidentally or intentionally exposed to high concentrations of HQ. Dermal exposure to HQ in humans is associated with vitiligo, a skin disease characterized by areas of depigmentation, and ochronosis, a condition of hyperpigmentation of the skin. HQ and benzoquinone have been shown to form DNA adducts *in vitro*, including in HL60 human bone marrow cells. Mutagenicity studies of HQ are generally negative in *S. typhimurium*. However, increased incidences of chromosomal aberrations, abnormal mitoses, formation of micronuclei, and sister chromatid exchange have been observed in other genotoxicity studies of HQ.

HQ is a metabolite of benzene and has been implicated in the carcinogenic response in humans exposed to benzene. Further, it has been hypothesized that exposure to HQ from sources such as dietary ingestion and over-the-counter (OTC) products may play a role in the etiology of leukemia in the general population. The International Agency for Research on Cancer (IARC) has determined that HQ is not classifiable as to its carcinogenicity to humans due to inadequate evidence in humans and limited evidence in animals. Oral administration of HQ to rodents in a 2-year bioassay conducted by the National Toxicology Program (NTP) resulted in some evidence of carcinogenicity. Male rats had a higher incidence of renal tubular cell adenomas, female rats had a higher incidence of mononuclear cell leukemia, and female mice had a higher incidence of hepatocellular adenomas or carcinomas following HQ treatment. There are no data describing chronic toxicity in animals following dermal administration of HQ. No overt

toxicity was detected in 14-day dermal studies conducted by the NTP in F344/N rats and B6C3F1 mice. However, the extent of absorption, bioactivation, and toxicity of HQ to the skin were not determined in these animals. Elsewhere, HQ metabolism was shown to be qualitatively similar in the F344/N rat following oral or dermal administration. However, toxicity may depend on quantitative differences in absorption and/or metabolism between routes of administration. Additional data are needed to determine the effect of dosing route on toxicity, particularly in a pigmented animal model, e.g. the B6C3F1 mouse.

Studies of potential reproductive and developmental toxicity for HQ have produced conflicting results. HQ was toxic to chick and rat embryos *in vitro*, with evidence of structural malformations in rats. Further, exposure to HQ has resulted in increased fetal resorption rates and adversely affected estrus cycles, spermatogenesis, and fertility in rats. However, other *in vivo* studies in rats, mice, or rabbits have resulted in negative results for HQ-mediated reproductive or developmental toxicity. Due to inconsistencies in these data and incomplete dermal toxicity data in animals described previously, additional toxicological characterization of HQ is needed to assess risk to humans following dermal exposure.

Key Issues:
The safety of OTC products containing HQ for depigmenting and lightening of skin is in doubt based on evidence of associated skin toxicity in humans and toxicity and carcinogenicity in experimental animals. The use of HQ in OTC products has been banned in the European Union and in Japan because of these concerns. Previously, OTC preparations containing ≤ 2% of HQ were classified as “generally recognized as safe and effective” (GRASE) in the U.S. However, due to the potential for toxicity, the FDA has withdrawn the ruling and requests additional toxicological data for HQ to aid in evaluating safety and developing regulatory policy for OTC products containing the chemical. Specifically, data are needed to assess reproductive toxicity in rodents and to determine potential toxicity and carcinogenicity in suitable animal models exposed to HQ by the dermal route.

Proposed Approach:
The main goal of this research program is to provide data for further toxicological evaluation of HQ. The specific aims are to:

1. Conduct comparative studies of the metabolism and disposition (ADME) of ^{14}C-labeled HQ in Sprague-Dawley rats and B6C3F1 mice by the oral and dermal routes. These studies will characterize the effect of the dosing route on internal dose and metabolism of HQ. The data are needed to support the design and interpretation of proposed dermal and oral toxicity studies in these specific rodent models. If needed, an abbreviated comparative study of the ADME of HQ in F344/N rats would be conducted to aid in correlating results from previous HQ studies in the F344/N rat with proposed studies in Sprague-Dawley rats.

2. Conduct a comprehensive reproductive toxicity study by the oral route in rats and mice utilizing the NTP Reproductive Assessment by Continuous Breeding (RACB) protocol. If indicated, supplemental developmental toxicity studies would be conducted.

3. Conduct toxicity and carcinogenicity studies of HQ by the dermal route in rodents. Sub-chronic studies would be conducted in the albino Sprague-Dawley rat and the pigmented B6C3F1 mouse to investigate dermal and systemic toxicity following topical application of HQ. The presence or lack of functioning melanocytes may affect dermal toxicity; therefore, results from these and the ADME studies will be used to determine the choice of rodent model(s) and the design for the proposed 2-year dermal toxicity and carcinogenicity studies.

Significance and Expected Outcome:
The proposed research program will provide necessary data for assessing the safety of OTC preparations used as skin-lightening agents. The data will be used by the FDA to develop regulatory policy for these products.

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